

SILICATE SLAG COMBINED WITH TEBUCONAZOLE IN MANAGEMENT OF BROWN EYE SPOT IN COFFEE

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ABSTRACT: The aim of this trial was to evaluate silicon absorption and the effect of silicate slag application combined with triazole fungicide in control of brown eye leaf spot under greenhouse conditions, using coffee seedlings of the cultivar Catuai Vermelho 144. A completely randomized 5x2 experimental design was used, with and without application of the fungicide tebuconazole, with eight replications. The mixtures (M) used on the soil were defined as based on the liming needs of the soil: M₁ - 0% de silicada slag (slag) and 100% limestone (lim); M₂ - 25% slag and 75% lim; M₃ - 50% slag and 50% lim; M₄ - 75% slag and 25% lim; M₅ - 100% slag, with or without application of the fungicide tebuconazole. The fungicide tebuconazole (0.5 g of a.i./L) was applied on coffee seedlings through five pair of leaves. Twenty-four hours after fungicide application, each coffee seedling was inoculated with a conidial suspension of 2.0×10^5 /mL. The severity and the defoliation caused by disease were evaluated every five days from 20 to 45 days after inoculation. With the severity and defoliation data, the area under the brown eye spot disease progress curve (AUCPBES) and the area under the defoliation curve (AUCPDEF) were calculated. The application of increasing doses of silicate slag on the soil did not reduce AUCPBES and AUCPDEF, and no increase in Si content in the root, stem or leaf was observed. Tebuconazole reduced AUCPBES and AUCPDEF.

Index terms: Silicon, control, *Coffea arabica*, *Cercospora coffeicola*.

ESCÓRIA SILICATADA COMBINADA COM TEBUCONAZOL NO MANEJO DA MANCHA DE OLHO PARDO DO CAFEIEIRO

RESUMO: Objetivou-se, neste ensaio avaliar a absorção de silício e o efeito da aplicação de escória silicatada combinada com fungicida triazol no controle da mancha de olho pardo em casa de vegetação, utilizando mudas de café da cultivar Catuai Vermelho 144. O experimento seguiu o delineamento inteiramente casualizado 5x2 com e sem aplicação do fungicida tebuconazole, utilizando oito repetições. As misturas (M) utilizadas no solo foram definidas com base na necessidade de calagem do solo: M₁-0% de escória silicatada (ESC) e 100% de calcário (CAL); M₂-25% ESC e 75% CAL; M₃-50% ESC e 50% CAL; M₄-75% ESC e CAL 25%; M₅ -100% ESC com ou sem aplicação de fungicida tebuconazole. O fungicida tebuconazole (0,5 g de ia / L) foi aplicado em mudas de café com cinco pares de folhas. Vinte e quatro horas após a aplicação do fungicida, cada muda de café foi inoculada com uma suspensão de conídios de $2,0 \times 10^5$ mL⁻¹. A severidade e a desfolha causada pela doença foram avaliadas a cada cinco dias dos 20 aos 45 dias após a inoculação. Com os dados de severidade e desfolha, foram calculadas a área abaixo da curva de progresso da severidade da mancha de olho pardo (AACPMP) e a área abaixo da curva de progresso da desfolha (AACPDEF). A aplicação de doses crescentes de escória silicatada no solo não reduziu AUCPBES e a AUCPDEF e nenhum aumento no teor de Si na raiz, caule ou folha foi observado. O tebuconazole reduziu AUCPBES e AUCPDEF.

Termos para indexação: Silício, controle, *Coffea arabica*, *Cercospora coffeicola*.

1 INTRODUCTION

The brown eye spot, caused by the fungus *Cercospora coffeicola* Berkeley & Cooke is one of the main coffee plant diseases, causing damage up to 30% in the yield of adult coffee plants, especially in terms of greater sunstroke. The brown eye spot occur naturally in the field, however, is more problematic in the greenhouse phase, causing intense defoliation especially when the plants are malnourished and under conditions of high relative moisture, in this case making the not viable seedlings for planting (CUSTÓDIO et al., 2011; POZZA et al., 2001; ZAMBOLIM; VALE; ZAMBOLIM, 2005).

The symptoms of the brown eye spot are stains of light brown to dark brown with a clear center and involved by a yellow halo, giving the aspect of an eye (ZAMBOLIM; VALE; ZAMBOLIM, 2005). The control of this disease is made specially with the use of fungicides, however the chemical control can result in the increase of production costs, intoxication farmers, contamination of the environment and risk in selection isolates of pathogens resistant to fungicides (KOLER, 1998; ZAMBOLIM; VENÂNCIO; OLIVEIRA, 2007). Therefore, there is a need to search for an alternative method in controlling the disease to be a part of an integrated management, which has as premise

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the use of one more strategy of disease control (ZAMBOLIM; VALE, 2003). Among the strategies of disease control in plants, the mineral nutrition is important, since it is possible to be manipulated, enabling its use as complement or alternative method in disease control (MARSCHNER, 1995). The silicon (Si) stands out for reducing the severity of important diseases in several cultures including dicotyledons although its effect is more pronounced in monocotyledons (AMARAL et al., 2008; BOTELHO et al., 2005, 2009, 2011; EPSTEIN, 1999; FAUTEUX et al., 2005; FRENCH-MONAR et al., 2010; RODRIGUES; DATNOFF, 2005; RODRIGUES et al., 2003).

The resistance of coffee plants to diseases can be increased with the equilibrated nutrition (POZZA et al., 2004b). Botelho et al. (2005) observed a reduction in the intensity of the coffee brown eye spot in coffee plants cultivated in a substrate that has received the application of calcium silicate and sodium silicate at doses of 0.5, 1 and 2 g kg⁻¹ of substrate. Similar results were observed by Pozza et al. (2004a) when 1 g CaSiO₃ Kg⁻¹ of substrate was applied. However, in coffee seedlings of cv. IAC 144 which had supplied silicon in nutrient solution no change was observed in epidemiological components of coffee rust (CARRÉ-MISSIO et al., 2009).

The use of calcium and magnesium silicates in the shape of slag silicates has been even more common in agriculture, seeking the correction of acidity, provide nutrients and reduce diseases (KORNDORFER; PEREIRA; CAMARGO, 2003; SOUSA; KORNDÖRFER; WANGEN, 2010). Studies show that the calcium one of the main nutrients found in these correctives can reduce the stain of coffee brown eye (GARCIA JÚNIOR et al., 2003). However the silicate slag still contains high levels of silicon, could be an alternative for the management of brown eye spot in coffee seedlings.

However, there are no studies regarding the absorption and control of diseases by Si, when applied in coffee seedlings, in the shape of slag and the interaction of this strategy as chemical control. Facing these factors, the objective of this paper was to evaluate the absorption of silicon and the effect of different doses of slag combined with the fungicide tebuconazole (triazole), on the severity of the brown eye spot in coffee seedlings.

2 MATERIAL AND METHODS

The experiment was conducted in the greenhouse in the period of September 10th to April 20th of 2008 in seedlings of coffee of the

cultivar Catuaí Vermelho 144, susceptible to the brown eye spot. The experimental design used was completely randomized design in the factorial scheme 5x2, using doses of slag, with or without the application of the fungicide tebuconazole, using eight repetitions. To define the quantity of slag to be applied, chemical analysis of the soil and the calculation of the need for liming (NC) based in the Recomendações para o uso de corretivos e fertilizantes em Minas Gerais -5^a Aproximação. After obtaining the NC, the slag was used (PRNT= 68%) and the limestone (PRNT= 95%) to define the quantity of correction has to be applied according to the following mixtures (M): M₁: 0% de slag e 100% de limestone (Lim); M₂: 25% slag and 75% Lim; M₃: 50% slag and 50% Lim; M₄: 75% slag and 25% Lim; M₅: 100% slag and 0% Lim the following quantities corresponding to M₁: 2.75 Kg/m³ de Lim; M₂: 0.8 slag and 2.1 Kg/m³ Lim; M₃: 1.38 slag and 1.59 Kg/m³ Lim; M₄: 2.49 slag and 0.69 Kg/m³ Lim; M₅: 3.16 Kg/m³ slag. The limestone was added to the mixture applied with the goal of standardizing the correction requirement of 100% and maintains the calcium similar for all treatments, ranging thus only silicon content added via silicate slag. The used slag consisted of 36% de CaO, 9% of MgO and 22.4% of SiO₂ (10.5% of Si).

The soil, Dystrophic Red-Yellow Latosol (LVAd), used as substrate was classified as argillaceous and presented a need for liming of 3.3 tons/2.000m³ of soil and possessed 0.1 cmolc/dm³ of the Ca²⁺; 0.0 cmolc/dm³ of the Mg²⁺; 45.3 mg/dm³ of the P; 15.0 mg/dm³ of the K and 6 mg de Si/Kg de soil. The soil received application of the corrections 15 days before plantings. The correction was homogenized to the soil and the moisture kept at field capacity. Next, placed in each 2L vases, a coffee seedling, when is in the matchstick phase was and maintained in greenhouse until the end of evaluations. Each vase of 2L corresponding to one experimental unit.

To obtain the conidia of *C. coffeicola* seeking the inoculation of plants, we used the methodology growth diphasic. Then, the suspension was calibrated for a concentration of 2.0x10⁵ conidia mL⁻¹. When the seedlings had five pairs of leaves, four months after transplanting, the fungicide tebuconazole (0.5g of i.a/L) was applied in the corresponding treatments, in the adaxial faces of the five pairs of younger leaves of the plants. After 24 hours of application with the fungicidal treatments, every plant was inoculated with the fungus in the abaxial and adaxial faces of the leaves with the help of a De Vilbiss n° 15

atomizer, then transferred to a fog chamber at 25 °C with continuous light for 12 hours. The evaluations were made every five days, from the 20th to the 45th day after inoculation. The severity was evaluated using a diagrammatic scale proposed by Oliveira et al. (2001). However, since the disease caused the defoliation of the seedlings, the severity decreases throughout the assessments. To correct this problem, the severity of the previous evaluation plus the current evaluation was used. With the data of severity and defoliation, was calculated the area under the curve of progress for severity (AACPMOP) and the area under the curve of defoliation (AACPDES) of the brown eye spot according to Shaner and Finner (1977).

After the end of the experiments, samples from the soil, leaves, stem and root from all of the experimental units received the same dose of slag even if they have not received the fungicide. The samples were collected to dry in an oven with forced air ventilation at 70 °C, until reaching constant weight and the rate of Si was determined according to the methodology proposed by Korndörfer, Pereira and Nolla (2004).

The variables AUCPBES and AUCPDES and Si contents in the soil, leaf, stem and root were submitted to variance analysis and verification of the assumptions of normality, homogeneity and independence of experimental error. Later were performed regression analysis based on the percentage of slag and the application of fungicide tebuconazole or not, coded as 1 and 0, respectively, whose coefficients of the largest model used were tested by Student's t test at 5% significance to the best fit model with all significant terms, as follows: $y_i = \beta_0 + \beta_1 s_i + \beta_2 s_i^2 + \beta_3 f_i + \beta_4 s_i f_i + e_i$; where:

y_i = observed value of the variable in the observation i ($i = 1, 2, 3, \dots, 80$);

s_i = percentual de slag na observação i (0, 25, 50, 75 e 100%);

f_i = application of the fungicide tebuconazole in observation i (0 = no fungicide and 1 = with fungicide);

β_0 = constant regression;

$\beta_1, \beta_2, \beta_3$ e β_4 = regression coefficients;

e_i = the regression error associated with the observed value y_i .

3 RESULTS AND DISCUSSION

The application of slag in doses recommended by NC increased ($P < 0,05$) the Si content in the soil in more than 200%, from 5.0 mg Si / kg soil in a lower percentage of slag (0% slag and 100% Lim) to 15.4 mg Si / kg soil in the

greatest percentage of slag (slag 100% and 0% CAL) (Figure 1). The average content of Si in the root of was $1.30\% \pm 0.16$, higher than that found in the stem ($0.37\% \pm 0.04$) and leaf ($0.27\% \pm 0.03$). However, despite the increase in Si content in the soil by increasing the percentage in the mixture of slag applied, there was no increase ($P > 0.05$) on leaves, stem and root of seedlings of coffee, when these plants were grown on this substrate.

Substrate that received application of 6 g of silicic acid per kg of soil increased the Si content for 68 mg dcm^{-3} ; however plants grown on this substrate showed no increase in silicon concentration in the leaf and no change in most epidemiological variables related to brown eye spot studied (BOTELHO et al., 2011). Botelho et al. (2005) worked with brown eye spot not observed accumulated of silicon in the leaves with the application of the dose at $1.26 \text{ g SiO}_2 \text{ Kg}^{-1}$. Coffee plants grown for 45 days in nutrient solution containing 2 mmol L^{-1} showed higher Si levels in roots however no difference was observed in leaf content, thus demonstrating the inefficiency of coffee plants to absorb this element (CARRÉ-MISSIO et al., 2009) similarly or observed in this trial.

No interaction was observed between doses of slag and application of fungicides and the only variable fungicide application were significant in the model. There was no reduction ($P > 0.05$) the AUCPBES and AUCPDES according to the percentage of slag applied (Figure 2), showing that the application of slag alone or in mixture with Limestone in doses recommended by the NC did not reduce the severity and defoliation caused by the brown eye spot. Similarly results was founded for Botelho et al. (2011) that no founded change in most epidemiological variables related to brown eye spot studied.

Different results were found by Botelho et al. (2005), where was observed reduction brown eye spot intensity applying sodium silicate how silicon source up to dose to $1.26 \text{ g SiO}_2 \text{ Kg}^{-1}$ of soil, but the authors did not correlate the amount of silicon in tissues with the intensity of the disease in this case is difficult conclude that the effect is due to silicon. Already Pozza et al. (2004b) observed that plants in the 'Novo mundo' and 'Catuai' cultivar growing in soil with application of 1 g of CaSiO_3 showed low disease severity and verify the presence of silicon in the leaf by scanning electron microscopy. Amaral et al. (2008) observed a reduction in the intensity of brown eye spot in seedlings and in plants in field that received application of potassium silicate foliar at dose 1.5 ml L^{-1} .

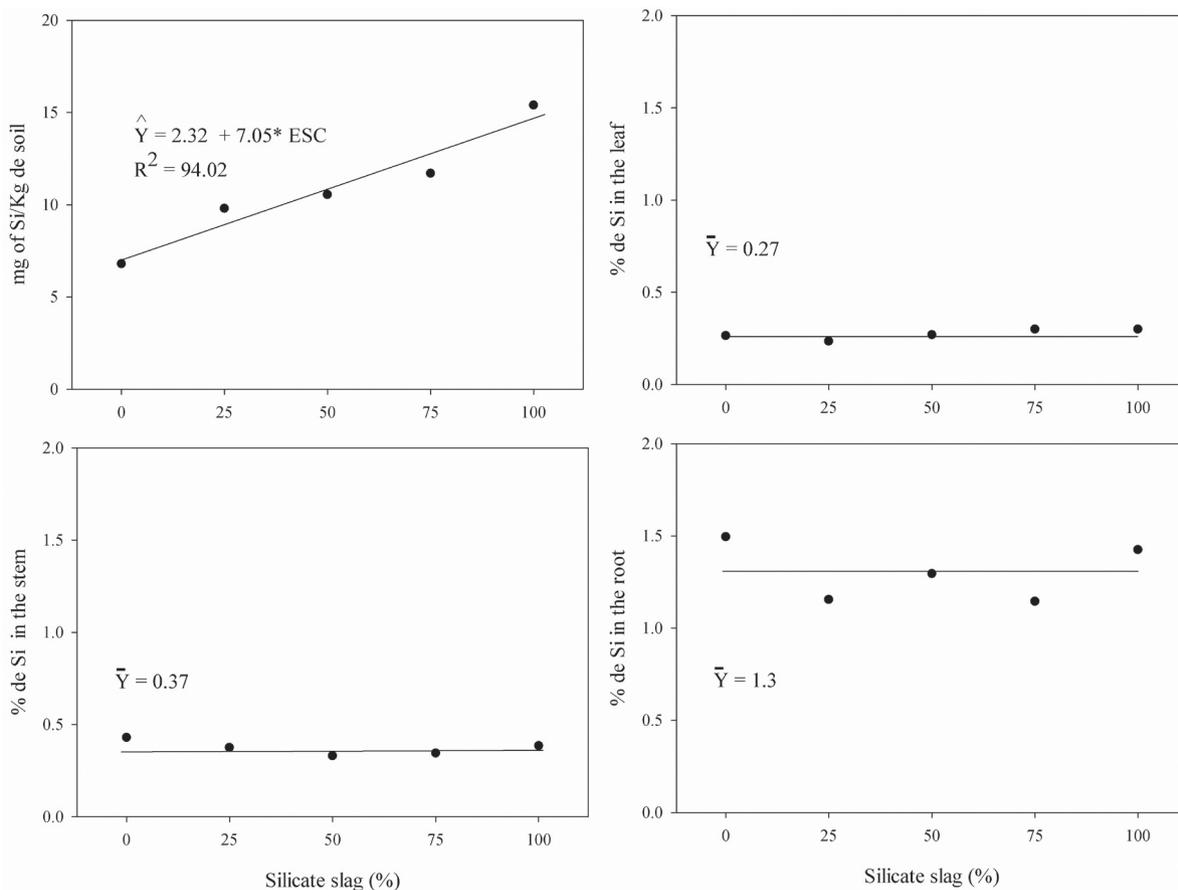


FIGURE 1 – Estimates of the amounts of Si in soil (mg/kg), leaf, stem and root (%) of seedlings of coffee as a function of the percentage of silicate slag.

* Significant by Student's t test ($P < 0.05$).

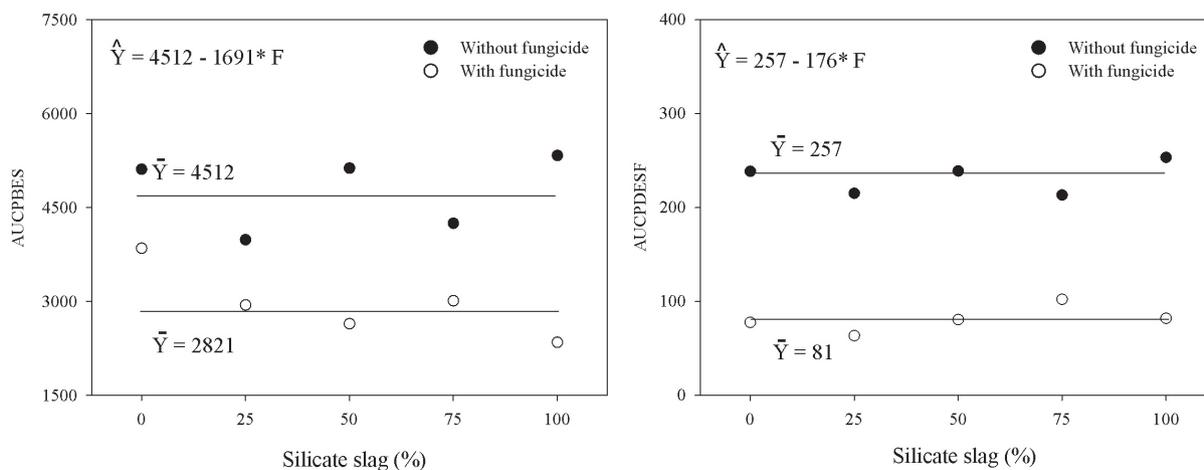


FIGURE 2 – Estimates of area under the curve of progress of the severity of brown eye spot (AUCPBES) and the area under the curve of defoliation (AUCPDEF) according to the percentage of slag silicate in the mixture with limestone and the application of fungicide tebuconazole (F = 0 indicates no fungicide and F = 1 indicates with fungicide).

* Significant by Student's t test ($P < 0.05$).

The silicon operates mainly forming the physical barrier formed by the deposition of that element below the cuticle and the wall of the epidermal cells preventing pathogen penetration (CHÉRIF; ASSELIN; BÉLANGER, 1994; EPSTEIN, 1999; KIM et al., 2002) or the potentiating of defense mechanisms (DATNOFF; RODRIGUES; SEEBOLD, 2007).

Thus, to ensure reduction in the intensity of diseases, silicon should be present at the site of infection or potentiate defense mechanisms. Therefore, this element need to be absorbed and accumulated in the site of infection. As the Si was not detected in greater quantities in leaves of coffee seedlings grown in substrate previously treated with slag, and the leaves are the organs infected by the fungus, there was no effect of Si on the disease. In studies with coffee seedlings grown in nutrient solution containing Si did not observe translocation of this element to the leaves, and no effect on leaf coffee rust (CARRÉ-MISSIO et al., 2009). The foliar application of Si is not resulted in increase of this element in coffee leaves as observed by Pereira et al. (2009), however Amaral et al. (2008) showed reduction brown eye spot with foliar application of silicon. Probably the silicon need not be absorbed but need to be at the site of infection.

The foliar application of tebuconazole reduced ($P < 0.05$) diseases severity and defoliation as shown by lower values of AUCPBES and AUCPDEF. There was a reduction of 37% of AUCPBES and 65% of AUCPDEF in units that received the application of the fungicide tebuconazole compared to those who have not received the application of the product. Since the strategy of chemical control with the use of triazole fungicides, the tebuconazole, is still an effective strategy for management of this disease, and recommended its use in nurseries of coffee seedlings in order to produce higher quality.

The mixtures of slag and Lim with percentages of slag ranging from 0 to 100% correction of the NC no increased the silicon level in different plant organs coffee and no effect on the severity and defoliation caused by brown eye spot was observed. As most soils have NC values and characteristics that resemble the soil used in this work, the use of slag in the substrate in order to maximize the control this disease in nurseries is not recommended, but the slag can be used to

others purposes. The limestone and slag can be used with the objective of providing the calcium to seedlings for reduce the spot brown eye (GARCIA JÚNIOR et al., 2003).

4 CONCLUSIONS

The coffee seedlings don't absorb the silicon.

The addition of silicate slag in the soil, in the quantity evaluated of this source, was not efficient in the control of brown eye spot under nursery conditions.

The use of the fungicide tebuconazole is still an appropriate strategy for the management of this disease.

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